

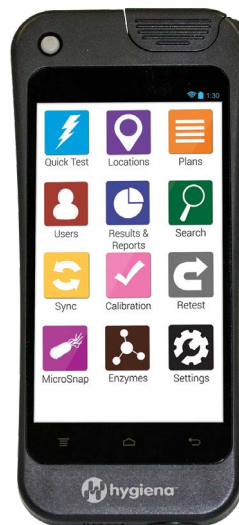
Comparing the Performance of ATP Hygiene Monitoring Systems

Hygiena™ vs Charm



ATP Hygiene Monitoring

ATP hygiene monitoring is a simple, rapid and quantitative testing method to verify cleaning effectiveness. For a surface to be verifiably clean, all food debris and other organic matter must be removed. Food debris, organic matter, and microorganisms contain ATP. Microorganisms are very tiny and individually contain only small amount of ATP. Thus, large numbers of microbes (~10,000) are required to be detectable by ATP test systems, which measure ATP residue in Relative Light Units (RLU). Systems are highly sensitive and can detect extremely low levels of ATP molecules, which means they can detect extremely small amounts of organic matter or food debris on surfaces. Effective cleaning removes both microbes and food residues. This means the lower the ATP reading is, the higher the cleaning standards are, resulting in a lower risk of microbial contamination.



What Has Changed?

Over the past 10 years, some systems have been re-designed, and some have received 3rd party certification by AOAC-RI under the *Performance Tested MethodsSM* Program.

Hygiena Changes

- Hygiena released the **EnSURE Touch™** to complement its **SystemSURE™ Plus** and **EnSURE™** luminometers.
- UltraSnap™** Surface ATP Test remains the same and is fully compatible with all three luminometers.
- UltraSnap** is an AOAC-validated method when used with **EnSURE** and **EnSURE Touch**.

Charm Changes

- Released **novaLUM II** to replace **novaLUM**.
- PocketSwab® Plus** ATP swab device remains the same.
- Neither Charm system has AOAC-certification.

Key Performance Characteristics of ATP Hygiene Systems

The critical performance characteristics of ATP hygiene monitoring systems are:

- Sensitivity** - the smallest amount of ATP and food residues detectable
- Consistency** - the variation of result from repeated tests of the same sample
- Accuracy** - the measured ATP value compared to the true value
- Precision** - the repeatability of the test to produce the same result

These parameters are determined using samples containing several different concentrations of ATP, including a sample without ATP. Ten replicates at each concentration level are tested. The data generated is used to calculate the limit of sensitivity, consistency, accuracy and precision.

Sensitivity

The table below shows the smallest amount of ATP detectable by each ATP hygiene monitoring system. Hygiena systems show a continual improvement over the past 10 years; whereas the Charm systems have remained the same. In fact, Hygiena's ATP system is 10x more sensitive than Charm's luminometer, **novaLUM II**. If greater sensitivity is required for high risk operations, then Hygiena's SuperSnap™ High-Sensitivity Surface ATP Test provides an additional 5-fold increase in sensitivity (not shown in table).

| Lowest amount of ATP (fmols) detected = greater sensitivity | Hygiena UltraSnap | | | Charm PocketSwab Plus | |
|---|-------------------|--------|-----------------|-----------------------|---------|
| | EnSURE Touch | EnSURE | SystemSURE Plus | novaLUM II | novaLUM |
| | <1.0 | 1.0 | 1.0 | 10.0 | 10.0 |

Key Factors Affecting Sensitivity

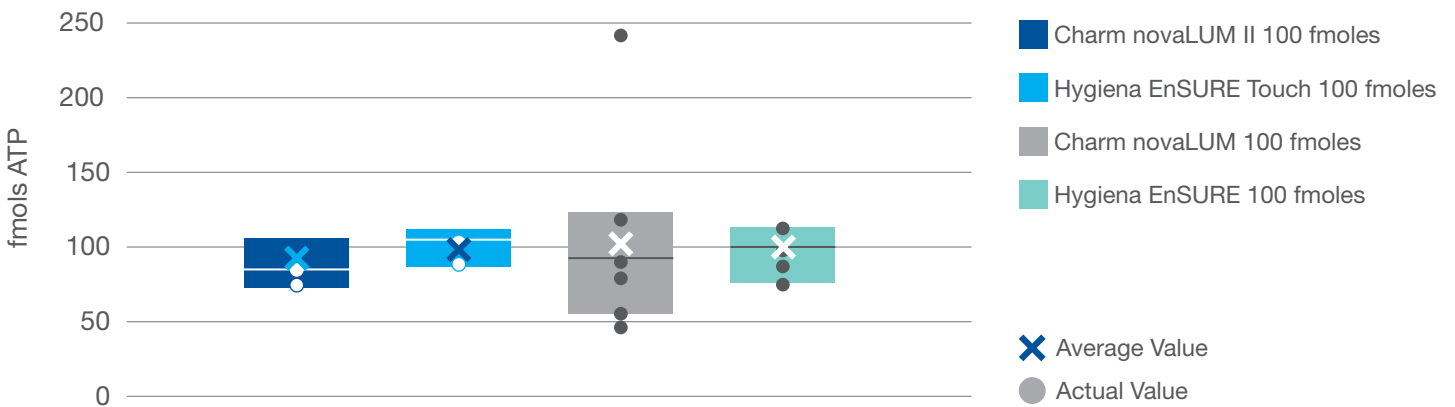
Each detection system will generate a response when there is no ATP in the sample. This is called background noise and is caused by impurities in the chemistry. If not removed, these impurities significantly affect the performance of the system. For freeze-dried reagents like that found in Charm's **PocketSwab Plus**, these impurities are locked in at the point of manufacture. Unlike Charm, Hygiena's liquid stable chemistry remains active and impurities are removed.

To compensate for background noise, Charm instruments have a built-in bias that does not display results at low RLU. While this may hide background noise, it limits the system's ability to detect low levels of ATP.

Hygiena's liquid stable chemistry reduces background noise and eliminates the need for a built-in biases. This means Hygiena's systems are able to detect ATP at lower levels. Hygiena's systems provide more reliable and sensitive measurements, particularly at low level detection required for cleaning verification.

At typical Pass / Fail threshold limits (e.g. 10 / 100 fmols of ATP), the graph below shows that the Charm's **PocketSwab Plus** is more variable (height of **grey** bar) regardless of using the **novaLUM** or **novaLUM II** luminometer. In contrast, Hygiena systems deliver the most consistent result closest to the expected value. Accordingly, Hygiena systems have greater precision and accuracy.

Comparison of accuracy and precision of Hygiena's **EnSURE** and **EnSURE Touch** versus Charm's **novaLUM** and **novaLUM II** measuring 100 femtomoles with US2020 and **PocketSwab Plus** ATP swab

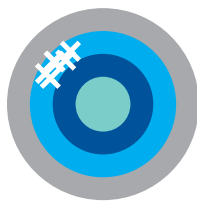


High Precision / High Accuracy

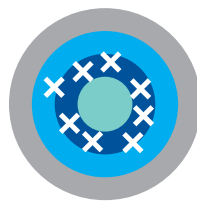


Hygiena
BEST

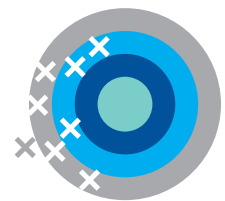
High Precision / Low Accuracy



Low Precision / High Accuracy



Low Precision / Low Accuracy



Charm
WORST

Detection of Food Residues and Microbes

Internal data shows Hygiena detected similar or smaller amounts of food residues directly added to swabs compared to Charm. In the earlier Siliker study, different foodstuffs and experimental design was used, but both systems showed similar sensitivities to food residues.

The AOAC study* for **EnSURE** and **EnSURE Touch** showed that both systems were able to detect bacteria and yeast. The smallest number of microbes detected by Hygiena systems was ~50,000 bacteria and 1,000 yeasts. This was similar to the Siliker study of 2010 that also showed Hygiena's **SystemSure Plus** system was more sensitive detecting microbes than Charm's **novaLUM** system.

The ATP surface cleaning verification test is not intended to be a surrogate bacteria test because it does not have the required sensitivity (typically 250/100cm² swab area).

There is no data available for **novaLUM II** because an AOAC study does not exist.

| Food residues added directly to swabs (customer comparison data 2018) | ATP System | | Food residues added directly to swabs (Siliker 2010) | ATP System | |
|---|----------------------|------------------|--|-------------------------|---------------|
| | Hygiena EnSURE Touch | Charm novaLUM II | | Hygiena SystemSURE Plus | Charm novaLUM |
| Resuspended milk powder | < 1 in 1,000 | < 1 in 1,000 | 2% low-fat milk | < 1 in 1,000 | 1 in 100 |
| Vegetable baby food | 1 in 100 | 1 in 5 | Bagged mixed salad | 1 in 1,000 | 1 in 1,000 |
| Chicken breast | 1 in 1,000 | 1 in 100 | Ground beef | 1 in 10,000 | 1 in 100 |

Third-party data provided by Siliker labs.

Summary

- Hygiena systems are the most sensitive, accurate and consistent. They are verified independently and have maintained best-in-class performance over the past 10 years.
- EnSURE and EnSURE touch are certified by the AOAC-RI *Performance Tested Methods*SM Program.
- Charm's **novaLUM** and **novaLUM II** are less sensitive and more variable than all Hygiena systems.
- Charm's **novaLUM II** has similar sensitivity to **novaLUM** but results are more variable.
- Charm systems are not certified by the AOAC-RI *Performance Tested Methods*SM Program.

*Data provided by Hygiena AOAC certificate #101803.

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PocketSwab™ Plus is a trademark of Charm.